REMARKS

Claims 1, 2, 4, 5, 7-20, 22, 23, 59, and 61 have been canceled without prejudice or disclaimer. Claims 62-91 have been added and therefore are pending in the present application. Claim 62 and 75 are supported by claims 1, 2, and 59 as originally filed, page 14, lines 13-14, and page 12, lines 5-12. Claims 63-74 and 76-91 are supported by the claims as originally filed.

It is respectfully submitted that the present amendment presents no new issues or new matter and places this case in condition for allowance. Reconsideration of the application in view of the above amendments and the following remarks is requested.

I. The Rejection of Claims 1, 2, 4, 5, 7-20, 22, 23, 59, and 61 under 35 U.S.C. 112

Claims 1, 2, 4, 5, 7-20, 22, 23, 59, and 61 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for the method wherein the integration of amplification units is selected for by reconstitution of a non-functional galactose utilization gene in a bacterial host cell, does not reasonably provide enablement for a method wherein the selection is made by another mechanism in another host cell. This rejection is respectfully traversed.

Applicants respectfully submit that one of ordinary skill in the art would be able to use other host cells and other mechanisms for selecting host cells. However, in order to advance prosecution, the newly presented claims are drawn to bacterial host cells, and to the reconstitution of genes of the host cell encoding at least one enzyme involved in the removal of UDP-galactose from the bacterial cell when the cell is grown in the presence of galactose or a galactose precursor.

For the foregoing reasons, Applicants submit that the claims overcome this rejection under 35 U.S.C. 112. Applicants respectfully request reconsideration and withdrawal of the rejection.

II. The Rejection of Claims 1, 2, 4, 5, 7-20, 22, 23, 59, and 61 under 35 U.S.C. 112

Claims 1, 2, 4, 5, 7-20, 22, 23, 59, and 61 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite. The Office provided several grounds for the rejection. Several grounds for the rejection are respectfully traversed. The remaining grounds are overcome by the newly presented claims.

First, the Office objected to the phrase "wherein a chromosomally integrated copy of the amplification unit is duplicated or multiplied on the host cell chromosome," recited in claim 1 becaus it is unclear how the amplification unit (henceforth AU) duplicat s or multiplies by itself

following integration. The Office also proposes three interpretations of the claim. This ground is respectfully traversed.

The specification on page 12, lines 29-33 clearly defines amplification units as follows:

> An amplification unit of the invention is a nucleotide sequence that can integrate into the chromosome of a host cell, whereupon it can increase in number of chromosomally integrated copies by duplication of multiplication. The unit comprises an expression cassette as defined herein comprising at least one copy of a gene of interest and an expressable copy of a chromosomal gene, as defined herein, of the host cell.

The ways an amplification unit may be amplified are well known in the art. An AU provides the means to achieve its own duplication on the chromosome when it is integrated into the chromosome of a host cell. In some cases, an AU carries a couple of flanking direct repeats which serve as recognition sequences for homologous recombination and duplication. In other instances the AU integrates in such a manner as to create flanking repeated sequences once it has integrated properly. In either case, after one copy of the AU is integrated into the chromosome, the two homologous sequences align and recombine with each other, which results in duplication of the AU on the chromosome.

Applicants enclose a copy of Janniere *et al.* (1985, Stable gene amplification in the chromosome of *Bacillus subtilis*. Gene, Vol. 40: 47-55), which describes a way in which an AU can be amplified. In Fig. 3 of the Janniere publication are shown different AU's indicated by double-arrows, some of which are made up of wholly integrated DNA, and others of integrated DNA as well as chromosomal DNA. As described therein, after the AU is integrated into the chromosome, a double crossover event occurs between the two homologous sequences, which results in a duplication of the AU on the chromosome.

Consequently, the skilled artisan would have no doubt whatsoever about how the AU of the present invention duplicates or multiplies by itself following integration.

The Office also stated that "since there is no prior discussion of introducing a first AU into the host cell, there must already be one in the chromosome...." This rejection is respectfully traversed.

The claims do not require the presence of an AU prior to integration of the AU on the chromosome. Rather, as described above, the AU provides the means to achieve its own duplication on the chromosome when it is integrated into the chromosome of a host cell.

Second, the Office obj cted to the phrase "s lecting a host cell comprising two or more chromosomally integrated copies of the AU," recited in claim 1 because "it is unclear if the method

is designed to integrate multiple copies of the AU at one time, or if the selection is meant to include a single integration ev nt into a cell that has already undergone a previous integration event. This is again related to the fact that there is no indication in the claim that a previous integration event had occurred with respect to the AU." This ground is respectfully traversed.

The claims do not require the integration of multiple copies of the AU at one time. As described above, the AU provides the means to achieve its own duplication on the chromosome when it is integrated into the chromosome of a host cell. Thus, it is only necessary to integrate one copy of the AU on the chromosome.

Third, the Office stated that in relating to step (f), "in order to increase the number of integrated copies of the AU in the host cell, one would have to first render a second chromosomal gene non-functional in the host cell, therefore step f) should refer back to step a) ... or [i]n the very least, step f) should refer back to step c)...." This ground is respectfully traversed.

As explained above, the AU provides the means to achieve its own duplication on the chromosome when it is integrated into the chromosome of a host cell. Thus, it is only necessary to integrate one copy of the AU on the chromosome. Thus, step f) properly refers to repeating cycles of steps d) and e).

Fourth, the Office objected to the phrase "with each repeat," recited in claim 1 because there is insufficient antecedent basis. The newly presented claims do not recite this phrase. Therefore, this ground for the rejection has been overcome.

Fifth, the Office objected to the term "preferably," recited in claims 18 and 22. The newly presented claims do not recite this term. Therefore, this ground for the rejection has been overcome.

Sixth, the Office objected to Claim 19 because it contradicts a limitation set forth in an independent claim.

Claim 1 has been rewritten as claim 75 to address this objection.

For the foregoing reasons, Applicants submit that the claims overcome this rejection under 35 U.S.C. 112. Applicants respectfully request reconsideration and withdrawal of the rejection.

III. The Rejection of Claim 2 under 35 U.S.C. 102

Claim 2 is rejected under 35 U.S.C. 102(b) as being anticipated by Adams *et al.* (U.S. Patent No. 5,435,730). This rejection is respectfully traversed.

Adams *et al.* disclose a recombinant DNA molecule comprising th *Streptomyces* gal operon galK gene; galE gene; galT gene; P1 promoter; P2 promoter; P2 promoter expression unit; P1 promoter regulated reglon; or the entire *Streptomyces* gal operon.

However, Adams *et al.* do not disclose any "gene amplification" or "amplification unit", as claimed herein.

For the foregoing reasons, Applicants submit that the claims overcome this rejection under 35 U.S.C. 102. Applicants respectfully request reconsideration and withdrawal of the rejection.

IV The Rejection of Claims 17 and 59 under the Doctrine of Obviousness-Type Double Patenting

Claim 59 is provisionally rejected under the doctrine of obviousness-type double patenting as being unpatentable over claim 67 of U.S. Application No. 09/928,847 (Jorgensen *et al.*). Claim 17 is provisionally rejected under the doctrine of obviousness-type double patenting as being unpatentable over claim 77 of U.S. Application No. 09/928,847 (Jorgensen *et al.*). These rejections are respectfully traversed.

Jorgensen *et al.* disclose and claim methods for producing a protein by cultivating a host cell comprising at least one copy of a gene of interest encoding the protein, which is integrated at one or more specific and well-defined locations on the genome. For example, where a DNA construct comprising two non-functional conditionally essential genes and two copies of the gene of interest is introduced into the host cell, one copy of the gene of interest will integrate adjacent to or overlapping with one of the non-functional conditionally essential genes, and the other copy of the gene of interest will integrate adjacent to or overlapping with the other non-functional conditionally essential gene, which results in restoring the functionality of both conditionally essential genes, which in turn renders the host cell selectable.

In contrast, the methods of the present invention involves methods of producing a polypeptide by cultivating a bacterial host cell comprising two or more amplified copies of a gene of interest encoding the polypeptide, wherein the gene of interest is contained within an "amplification unit". This is done by integrating the amplification unit into any location of the chromosome of a host cell, growing the host cell under conditions conducive for duplication of the amplification unit, and selecting a host cell comprising duplicated copies of the unit on the chromosome, whereby the gene of interest will also have been duplicated. The growth and selection steps may be repeated to achieve multiple duplication events, leading to an increasing number of copi s of the gen of interest on the chromosome. Thus, the host cell produced by the

methods of the present invention contains a number of copies of a gene of interest, which are located adjacent to each oth r on the genome of the cell.

Thus, the host cells of the present invention are patentably distinct from the host cells disclosed in Jorgensen *et al.*

For the foregoing reasons, Applicants submit that the claims overcome these rejections under the doctrine of obviousness-type double patenting. Applicants respectfully request reconsideration and withdrawal of the rejections.

V. Conclusion

In view of the above, it is respectfully submitted that all claims are in condition for allowance. Early action to that end is respectfully requested. The Examiner is hereby invited to contact the undersigned by telephone if there are any questions concerning this amendment or application.

Respectfully submitted,

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